

Influence of Various Persistent Chlorinated Insecticides on the Macro and Micro Element Constituents of Zea Mays and Phaseolus Vulgaris Growing in Soil Containing Various Amounts of These Materials*

by H. COLE, D. MACKENZIE, C. B. SMITH, and E. L. BERGMAN

Pesticide Research Laboratory

Departments of Plant Pathology and Horticulture

Pennsylvania State University, University Park, Pennsylvania

Introduction

Intensive research efforts by many groups and individuals have been devoted to understanding the influence of pesticides in the ecosystem. These efforts have dealt with the effects of pesticides on man, birds, wild mammals, fish, "beneficial" insects, and other non-target organisms. This work has been summarized by Dustman and Stickel (1966, 2) and others.

However, one group of organisms, the higher plants, has been almost completely ignored in this research effort. This is especially true in the case of many insecticides and fungicides which are thought to have no "phyto-active" properties.

Lichtenstein (1965, 5) has shown that certain of the chlorinated hydrocarbon insecticides including aldrin, heptachlor and others are extremely persistent in soils and remain for long periods after application. Lichtenstein and Schulz (1960, 4) and Wheeler, Mumma and Frear (1965, 6) have shown that these materials may be absorbed by plants and distributed throughout the

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above-ground tissues. If effects on plants were to occur from the use of these materials, the effects might remain for as long as the material was in contact with the plant. These effects would not be restricted to areas of intensive crop production but could occur in marshland or other natural vegetation areas where pest control applications might be made. With the present interest in metallic element nutrition and its role in human health a knowledge of the effects of pesticides on the micro element constituents of crop plants is essential.

Because of the human nutrition aspects as well as the need for short term experiments, the crop plants, corn Zea mays var. indentata (Sturtev.) Bailey hybrid Ohio 43 x Pa 70 single cross, and beans Phaseolus vulgaris L. cultivar Bountiful, which can be easily grown under controlled conditions were chosen for study.

Experimental Methods

The test plants were grown in a greenhouse in glazed ceramic containers containing a 3:1:1 mixture of Hagerstown silty clay loam, peat, and fine sand. The containers had 20 mm openings in the bottom which were covered with a 1/16 inch layer of porous fiber glass cloth to allow drainage without soil loss.

The soil was steam treated at 100°C for one hour. Soil moisture content at the end of the steam treatment approximated field capacity. The soil was then allowed to cool to room temperature, thoroughly mixed to avoid treatment position effects, and the test chemicals were added to the soil. All chemicals

were mixed with soil on a "chemical weight/soil weight at field capacity" basis to achieve the desired ppm concentration. Because of the small quantities needed, each chemical was mixed with sufficient water to make 50 ml volume and this solution was mixed with 30 g of "Vermiculite". The Vermiculite pesticide mixture was then blended with the soil. In the control series 30 g of Vermiculite + 50 ml water was added to the soil. All chemicals used were commercial formulations with the quantities computed on the basis of active ingredient.

The materials and formulations tested included DDT 5% wettable, heptachlor 2 lbs/gal liquid, chlordane 4 lbs/gal liquid, and endrin 1.6 lbs/gal liquid. Dosages used for each chemical were 0, 1, 10, and 100 ppm. Immediately after the chemical was mixed with the soil, the containers which had been previously steam treated, were filled with 9 lbs of the soil-pesticide mixture per container. They were then seeded with 8 bean or 8 corn seeds per pot which were thinned to 4 plants per pot soon after emergence. The seeds were free of any chemical treatment. No supplemental plant nutrients were added during the duration of each experiment. The plants were watered by adding equal amounts to each container. The levels added were such that slight drainage occurred from each container after watering. The intervals were sufficiently close so that any plant size differences with corresponding water extraction rate differences would not significantly influence soil moisture levels among containers.

This method of watering was evaluated (by the authors) previously, with various statistical procedures, on both wet and dry weights of corn plants in the greenhouse. No differential effects due to soil moisture could be detected. Each treatment was replicated six times (6 containers) for each plant species, pesticide treatment, and treatment dosage. The containers were randomly redistributed on the greenhouse bench at 7 day intervals to avoid position effects. Series A, DDT, was planted 10/18/66 and maintained in a greenhouse at 68-82°F, Series B, heptachlor, was planted 11/9/66 and maintained at 64-73°F, Series C, chlordane, was planted 1/2/67 and maintained at 72-78°F, and Series D, endrin was planted 2/1/67 and maintained at 66-72°F. No supplemental lighting was provided. The experiments were planned so that chemicals, plant species, plant age at harvest, and dosage were part of an overall factorial design suitable for statistical treatment.

Care was taken to avoid contamination of the plants or soil with plant pathogens. No plant pathogens or diseases could be detected by visual observation or pathogen isolation procedures on the plant roots or foliage.

For each series, 3 replicates were harvested at the end of 4 weeks growth. The above ground portions of the plant were weighed, fresh weights recorded, and then dried in an oven at 140°F for 48 hours. At the end of 8 weeks growth the remaining 3 replicates were harvested and treated in a similar manner. All leaf and stem tissues from a replicate were ground together in a Wiley mill

to pass a 40 mesh screen. The Wiley mill had been specially altered to prevent trace element contamination. All samples were analyzed for 12 elements. A semi-micro Kjeldahl procedure was used in determining nitrogen. An emission spectrometric procedure was employed to determine simultaneously phosphorous (P), potassium (K), calcium (Ca), magnesium (Mg), manganese (Mn), iron (Fe), copper (Cu), boron (B), aluminum (Al), strontium (Sr), and Zinc (Zn). The procedures and equipment employed have been presented in detail by Baker et al (1964, 1). The spectrometric data conversion, data transformation and analyses of variance¹ were accomplished with Fortran programs on the IBM 7074 computer at The Pennsylvania State University Computation Center.

Experimental Results

DDT at 1 and 10 ppm significantly decreased the fresh weight of corn and bean plants after 8 weeks growth while at the 100 ppm level growth was not significantly different from the check.

Heptachlor, 1 ppm, decreased the fresh weight of corn and bean plants while 10 and 100 ppm increased their weight after both 4 and 8 weeks growth.

Endrin after 8 weeks at 100 ppm decreased the weight of corn plants, and at 10 and 100 ppm decreased the weight of bean plants.

Chlordane did not significantly affect the weight of corn or bean plants (Table 1).

¹ The analyses of variance and LSD for the plants' fresh weight were computed by hand.

TABLE 1
INFLUENCE OF CERTAIN PERSISTENT INSECTICIDES ON FRESH WEIGHT OF CORN AND
BEAN PLANTS GROWING IN STEAM TREATED SOIL CONTAINING THESE MATERIALS

Plant Species and Concentra- tion of Chemical in ppm	Fresh Weight ¹ of Plants in Grams									
	DDT		heptachlor		chlordane		endrin			
	4 weeks	8 weeks	4 weeks	8 weeks	4 weeks	8 weeks	4 weeks	8 weeks	4 weeks	8 weeks
Corn 0	6.5	19.2	5.4	10.7	7.0	20.1	4.1	27.1		
1	6.6	17.6*	4.2*	8.9*	6.7	20.7	3.8	27.8		
10	8.6*	17.9	6.2	17.2*	7.3	22.1	4.0	24.8		
100	9.0*	19.4	8.3*	17.4*	7.5	18.4	4.2	23.1*		
LSD 05	1.1	1.4	1.1	1.8	1.2	2.9	1.3	2.7		
Beans 0	7.2	9.7	6.2	8.5	7.8	13.1	7.0	18.8		
1	7.2	8.3*	4.1*	7.4	8.3	12.6	6.0	18.0		
10	7.9	6.7*	5.7	10.5*	6.5	12.7	6.4	12.8*		
100	7.9	10.3	8.1*	15.0*	7.6	10.6	6.0	12.4*		
LSD 05	1.0	1.3	1.1	1.5	2.5	3.7	1.4	3.1		

¹ Means of 3 replications.

* Significantly different from the check at the .05 probability level when weights subjected to "analysis of variance."

All materials tested significantly affected both macro and micro element levels of above-ground tissues of corn and bean after 4 and 8 weeks growth. In general element level shifts with beans differed from corn and the changes after 4 weeks were different from those after 8 weeks.

With DDT, manganese (Mn) was most significantly affected. In addition changes in N, P, K, Ca, Mg, Fe, Cu, B, Al, Sr, and Zn were noted depending on the plant species and plant age at analysis. With corn plants increasing levels of DDT increased Zn while with bean increasing levels of DDT decreased Zn (Table 2).

Heptachlor most significantly affected Mn with changes in N, P, K, Ca, Mg, Fe, Cu, B, Al, Sr, and Zn also occurring. In general for both plant species and ages N levels decreased as heptachlor levels increased. Zn Levels increased with increasing heptachlor after 8 weeks growth for both corn and beans (Table 3).

Chlordane had less effect on element levels than DDT or heptachlor. Mn levels were shifted in only a few instances. No changes in Ca were noted in any instance. N, P, K, Mg, Fe, Cu, B, Al, Sr, and Zn were affected to lesser extents. Zn levels increased with increasing chlordane levels in beans after both 4 and 8 weeks. (Table 4)

Endrin resulted in changes in P, K, Mg, Mn, Fe, Cu, B, and Zn levels. No significant changes occurred in Ca, Al, and Sr levels with endrin. A trend was noted that with corn after both 4 and 8 weeks N and Cu levels decreased with increasing endrin. (Table 5)

TABLE 2
INFLUENCE OF DDT ON MACRO AND MICRO ELEMENT CONSTITUENTS¹
OF ABOVE GROUND PORTIONS OF CORN* AND BEAN PLANTS

Corn		Percent of Dry Weight										PPM of Dry Weight				
Plant age	Chemical concentration	N	P	K	Ca	Mg	Mn	Fe	Cu	B	Al	Sr	Zn			
4 weeks	0	4.48	0.23	5.96	1.30	0.31	619	198	9.7	17	236	19	66			
	1	4.67	0.21*	5.53*	1.34	0.30	961*	138	8.3*	16	135*	21*	56			
	10	4.39	0.23	5.84	1.16	0.29	951*	150	9.1	16	149	19	70			
	100	4.53	0.31*	5.76	1.15	0.33	605	137	12.6*	15	114*	22*	84*			
8 weeks	0	3.17	0.20	5.73	1.35	0.37	350	133	8.9	18	109	21	54			
	1	3.17	0.20	5.62	1.51	0.34*	664*	162	9.7	23*	147	22	44			
	10	3.15	0.19	6.16*	1.33	0.30*	609*	132	9.7	17	101	21	45			
	100	2.21	0.18	4.60*	0.94*	0.34*	452*	115	9.9*	17	96	18*	60			
Bean																
4 weeks	0	5.29	0.14	2.87	1.99	0.28	333	291	7.3	24	377	26	69			
	1	6.00*	0.18*	3.16	1.64*	0.24*	399*	211*	6.3	28*	296	24*	52*			
	10	5.86*	0.20*	3.95*	1.69*	0.30	464*	274	7.4	29*	376	25	46*			
	100	4.85*	0.18*	3.28*	2.00	0.32*	541*	202*	7.4	35*	234*	30*	41*			
8 weeks	0	3.95	0.16	2.62	2.35	0.28	330	210	7.1	28	177	29	55			
	1	4.53*	0.16	2.49	1.90*	0.23*	292	178	6.3	23*	190	26*	44			
	10	5.13*	0.15	2.64	1.90*	0.26	279	228	6.4	24*	279*	27*	34*			
	100	4.24	0.15	2.59	2.05*	0.30	512*	190	6.4	17*	201	31*	47			

¹ Means of 3 replications.

* Significantly different from the check at the .05 probability level when data subjected to "analysis of variance."

TABLE 3
INFLUENCE OF HEPTACHLOR ON MACRO AND MICRO ELEMENT CONSTITUENTS¹
OF ABOVE GROUND PORTIONS OF CORN AND BEAN PLANTS

Corn Plant age	Chemical concentration	Percent of Dry Weight					PPM of Dry Weight						
		N	P	K	Ca	Mg	Mn	Fe	Cu	B	Al	Sr	Zn
4 weeks	0	5.08	0.30	5.20	1.39	0.35	784	217	9.7	24	198	16	67
	1	4.68	0.23*	5.24	1.51	0.41*	423*	176	9.3	24	151	18*	53
	10	4.61	0.25*	5.22	1.35	0.39*	445*	186	9.7	21	177	16	61
	100	4.49*	0.29	5.13	1.43	0.40*	339*	187	10.8*	18*	162	17	76
8 weeks	0	4.32	0.17	5.01	1.59	0.38	767	173	8.3	27	145	18	46
	1	3.90	0.17	5.60*	1.64	0.41*	638*	170	7.9	23*	145	20*	45
	10	3.78*	0.22*	5.49*	1.41	0.63*	336*	152	9.4	24	108	19	59
	100	3.14*	0.21	4.34	1.49	0.55*	252*	158	9.1	17*	129	20*	64*
Bean 4 weeks	0	7.26	0.33	4.89	1.97	0.40	490	500	10.5	41	500	24	83
	1	7.83	0.33	4.20*	2.69*	0.37*	372*	420*	10.5	35*	402*	28*	68
	10	6.88	0.26*	4.02*	2.00*	0.37*	342*	384*	8.9*	32*	388*	25	63*
	100	6.42*	0.29*	4.20*	2.29*	0.46*	479	500	10.5	50*	500	27*	100*
8 weeks	0	7.25	0.22	3.31	2.10	0.33	212	497	7.3	27	705	24	55
	1	6.13*	0.19	3.07	2.62*	0.38*	292*	391*	7.4	26	447*	29*	37*
	10	5.02*	0.23	3.36	2.32	0.45*	405*	722*	7.8	40*	953*	29*	74*
	100	4.99*	0.26*	3.28	2.24	0.45*	423*	584*	8.2	43*	679	30*	89*

¹ Means of 3 replications.

* Significantly different from the check at the .05 probability level when data subjected to "analysis of variance."

TABLE 4
INFLUENCE OF CHLORDANE ON MACRO AND MICRO ELEMENT CONSTITUENTS¹
OF ABOVE GROUND PORTIONS OF CORN AND BEAN PLANTS

Corn Plant age	Chemical concentration	Percent of Dry Weight					PPM of Dry Weight						
		N	P	K	Ca	Mg	Mn	Fe	Cu	B	Al	Sr	Zn
4 weeks	0	5.05	0.30	5.64	1.48	0.30	463	211	9.9	17	185	17	78
	1	4.98	0.24*	5.80	1.46	0.29	424	198	8.3*	17	164	17	77
	10	5.27	0.32	6.12*	1.38	0.32	468	271*	12.3*	22*	291*	16	81
	100	4.81	0.26*	6.05*	1.40	0.32	306	218	9.1	17	210	16	79
8 weeks	0	2.53	0.14	4.20	1.05	0.34	281	100	6.1	14	88	11	47
	1	2.85	0.15	5.64*	1.19	0.29*	292	122	8.0*	16	99	14*	60
	10	2.78	0.14	5.38*	1.06	0.30*	255	96	6.6	15	70	12	51
	100	2.70	0.14	5.38*	1.24	0.30*	281	102	7.9*	18*	75	14*	56
<u>Bean</u>													
4 weeks	0	5.78	0.20	3.62	2.17	0.29	260	232	7.6	26	228	23	49
	1	6.31*	0.22	3.30	2.32	0.29	296	389*	7.5	26	440*	22	60
	10	6.33*	0.23	4.05*	2.26	0.33*	340*	401*	9.3*	25	437*	24	65
	100	6.23	0.24*	3.65	1.99	0.33*	351*	266	8.2	28	257	22	80*
8 weeks	0	3.83	0.17	2.06	1.86	0.25	275	207	5.1	25	217	19	45
	1	4.11	0.15	2.25	1.96	0.26	285	164	5.5	22	140	21	53
	10	3.94	0.16	2.23	1.87	0.26	248	133*	5.8	21*	82*	21	47
	100	4.46*	0.20	2.58*	1.87	0.29*	350*	170	6.4*	26	122	20	69*

¹ Means of 3 replications.

* Significantly different from the check at the .05 probability level when data subjected to "analysis of variance."

TABLE 5
INFLUENCE OF ENDRIN ON MACRO AND MICRO ELEMENT CONSTITUENTS¹
OF ABOVE PORTIONS OF CORN AND BEAN PLANTS

<u>Corn</u>															
<u>Plant</u> <u>age</u>	<u>Chemical</u> <u>concentration</u>	N	P	K	Ca	Mg	Mn	Fe	Cu	B	Al	Sr	Zn		
4 weeks	0	6.02	0.88	6.80	0.80	0.29	434	185	13.4	19	121	11	117		
	1	5.68	0.71*	5.84	0.87	0.31	454*	205	11.1*	25*	166	11	83*		
	10	5.67	0.88	6.30*	0.89	0.33*	533*	242*	12.8	21	137	11	91*		
	100	5.64	0.84	6.08	0.94	0.32	460	171	10.9*	18	117	12	96*		
8 weeks	0	3.45	0.54	4.71	0.73	0.27	362	119	8.5	15	74	9	65		
	1	3.43	0.44*	5.00	0.75	0.25	331	117	8.5	16	63	10	40*		
	10	3.20	0.48*	5.53	0.68	0.26	350	120	7.9	16	78	10	49		
	100	3.15	0.46*	4.87	0.84	0.30*	318	124	7.6*	15	78	11*	51		
<u>Bean</u>															
4 weeks	0	6.25	0.65	5.40	1.82	0.40	513	257	10.6	41	205	23	96		
	1	6.25	0.45*	5.00*	1.73	0.37*	499	224	9.1*	33*	171	22	75*		
	10	6.45	0.50*	4.95*	1.83	0.40	592*	275	10.0	35*	210	22	94		
	100	6.09	0.60*	4.89*	1.94	0.38	574	259	11.2	39	186	22	79*		
8 weeks	0	3.92	0.54	3.82	1.74	0.36	523	233	10.5	59	132	22	76		
	1	3.28*	0.36*	2.95*	1.98	0.36	495	244	7.7*	38*	177	21	51*		
	10	3.83	0.38*	3.90	1.52	0.32*	495	305*	8.2*	38*	198	20*	62		
	100	4.24	0.47*	3.93	1.88	0.37	558	234	8.2*	43*	119	22	69		

¹ Means of 3 replications.

* Significantly different from the check at the .05 probability level when data subjected to analysis of variance".

Discussion

It is believed that the experiments reported here have demonstrated that under the conditions of the tests the pesticides used significantly affected plant growth as evidenced by changes in weight as well as micro and macro element levels in above-ground tissues.

The changes induced by these materials at the 1 ppm level are of particular interest because of possible occurrence of similar levels outdoors in soils as well as on foliages from commercial use of these materials.

Ko (1967, 3) has just reported that certain soil fungi accumulate DDT and dieldrin in their hyphae in far greater amounts than is present in the surrounding soil and that certain soil bacteria and actinomycetes concentrate dieldrin in large amounts. These organisms could be expected to occur in association with plant roots.

In the present study the initial effects of soil microorganism-pesticide interaction were eliminated by steam treatment of the soil and containers prior to the addition of chemicals. It is recognized that spore forming bacteria would survive 212°F steam treatment; and fungi, bacteria and other organisms that were present on seed, or in greenhouse air and dust would re-colonize the steam treated soil at varying rates. The microorganism effects would be anticipated to be less than if untreated field soil were used.

Thus, the numbers and kinds of microorganisms present and their activities remain an unaccountable variable in this study, although it would be assumed that kinds of organisms and numbers remaining in soil after steam treatment and entering soil from greenhouse dust and air would be relatively constant for all the experiments. However this variable may be important in comparison of this work with any subsequent investigations under different conditions. It is felt that these introductory studies point up the need for definitive experiments with persistent insecticides to clarify in detail the mechanisms involved in eliciting growth response, the role of soil microorganisms in growth responses, and to determine if such effects occur under field conditions from commercial applications or drift from commercial applications.

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